

Martin Godfrey reports his findings from a recent study of *Cephaloziella* chromosomes.

Some chromosome counts for the genus *Cephaloziella*

Early in 2011, Des Callaghan asked me if I would do chromosome counts on a number of *Cephaloziella* species which he was studying. There are only a few published chromosome counts for this genus (Smith, 1996; Paton, 1999), so this was an opportunity to break new ground as well as confirm the published counts for *C. massalongi* and *C. nicholsonii* (Newton in Paton, 1984).

Methods

Martha Newton has published an excellent guide to bryophyte chromosomes (Newton, 1989) which formed the basis for this project. All of the counts were done using mitotic preparations of growing shoot tips but, as on previous occasions, I found that I needed to experiment a bit to achieve satisfactory results.

The living specimens were placed in small jars, liberally sprayed with water, sealed and allowed to grow for at least 2 weeks to ensure that there were plenty of growing shoot tips. The tips were harvested by cutting 5–10 mm lengths of shoots which were fixed overnight in 3:1 absolute alcohol/glacial acetic acid. My main departure from Newton was to stain the fixed shoots for 3–4 hours prior to the excision and squashing

△ *C. stellulifera*. D. Callaghan

of the growing tips as I have found that this gives more consistent results. I also found that with *Cephaloziella* I got better staining using aceto-orcein rather than lacto-propionic orcein. After staining, around 0.5 mm of the growing tip was excised in a drop of 45% acetic acid and squashed in the usual way. As the chromosomes were rather small, counts were made using a ×20 eyepiece with an oil immersion lens – in some cases a green filter was used to improve contrast.

Getting decent chromosome figures in *Cephaloziella* proved surprisingly difficult, especially in the case of *C. divaricata* which took many attempts. To check my technique I grew some *Cephalozia bicuspidata* alongside the *C. divaricata*, fixed and treated it in the same way and got excellent chromosome preparations, so it appears there is something about *Cephaloziella* which makes this genus rather intractable to standard techniques.

Results

Good counts were obtained for all of the species provided, and the results are summarized in Table 1. Not surprisingly, the new counts reflected what seems to be the underlying chromosome number for the genus, $n=18$, and it was pleasing

to confirm the known counts. In particular, finding that $n=36$ for the Welsh *C. nicholsonii* gives added support to the view (unpublished) that this species may well have arisen as a diploid of *C. massalongi*.

Duplicates of the specimens have been sent to RBGE and NHM for DNA sequencing studies. The vouchers have been retained in the personal herbarium of Des Callaghan.

Concluding remarks

I would strongly recommend that if you have not tried this very useful technique, you should do so. No special equipment beyond that typically owned by an active bryologist is needed and, whilst a bit of experimentation, practice and patience is required to get decent results, that

can be said of most bryological techniques. Obtaining halfway decent chromosome figures is probably simpler than cutting reasonable leaf sections for a beginner.

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References

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- Paton, J.A. (1999). *The Liverwort Flora of the British Isles*. Colchester: Harley Books.
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Table 1. Chromosome count results

*All specimens were collected and determined by Des Callaghan.

| Specimen no. | Taxon | Habitat | Site | Date collected | V.-c. | Altitude (asl) | Chromosome count |
|--------------|------------------------|--|------------------------|----------------|-------|----------------|------------------|
| 1295 | <i>C. divaricata</i> | On old mine spoil beside stream in open | Hermon Copper Bog SSSI | 23.02.11 | 48 | 150 | 18 (new count) |
| 1294 | <i>C. hampeana</i> | Thin humic soil over old mine spoil in open | Hermon Copper Bog SSSI | 23.02.11 | 48 | 150 | 18 (new count) |
| 1259 | <i>C. massalongi</i> | Partially worked metal ore seam at mouth of old copper mine | Figra Mine | 15.01.11 | 48 | 230 | 18 |
| 1281 | <i>C. massalongi</i> | On base of old mine building wall beside small stream in open mixed woodland | Pant-y-wrach | 04.02.11 | 48 | 40 | 18 |
| 1302 | <i>C. massalongi</i> | On soil deep under overhang of stream bank | Hermon Copper Bog SSSI | 23.02.11 | 48 | 120 | 18 |
| 1282 | <i>C. massalongi</i> | On old mine spoil of leat bank in mixed woodland | Coed Aberglaslyn | 05.02.11 | 49 | 60 | 18 |
| 1279 | <i>C. nicholsonii</i> | On old copper mine spoil on north-facing moorland slope | Drws-y-coed | 06.02.11 | 49 | 160 | 36 |
| 1284 | <i>C. rubella</i> | On stream bank beside old copper mine in mixed woodland | Pant-y-wrach | 04.02.11 | 48 | 40 | 18 (new count) |
| 1260 | <i>C. stellulifera</i> | Open area of flat spoil in old copper mine | Figra Mine | 15.01.11 | 48 | 230 | 18 (new count) |
| 1280 | <i>C. stellulifera</i> | On old copper mine spoil on north-facing moorland slope | Drws-y-coed | 06.02.11 | 49 | 160 | 18 (new count) |